

GENETICS, ENVIRONMENT, AND SUBSPECIES DIFFERENCES:
THE CASE OF *POLITES SABULETI*
(Lepidoptera: Hesperiiidae)

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ABSTRACT.—*Polites sabuleti* is an example of an insect having a univoltine, monophenic high-elevation subspecies and a multivoltine lowland one that produces similar phenotypes only in cold weather. When reared under conditions that induce the warm-weather phenotype in lowland stocks, the montane subspecies *P. s. tecumseh* continues to produce its usual phenotype, indicating that it has become genetically fixed.

One variant of the persistent "genetics-environment" duality in biology concerns the nature of subspecies differences. The problem, as it applies to butterflies, was well summarized in Klots's (1951) discussion of geographic variation:

To what degree much of the recorded geographic variation is a matter of temperature and humidity differences is something which we can only infer. In *Papilio glaucus* . . . spring specimens tend to be small and pale. . . . As we go northward we find that in central Canada, where there is only one generation a year, the whole population looks similarly small and pale. In Canada this population has been named as a geographic subspecies, "*canadensis*," i.e. a part of the species limited to a certain area and showing distinctive characteristics. The temptation is strong to attribute the whole thing to lowered temperatures alone. But suppose we brought a batch of eggs of *canadensis* down to Florida, and reared the butterflies in the conditions under which the very large, richly colored subspecies *australis* develops there. Would our *canadensis* eggs develop as *australis* . . . or would they develop into the same small pale specimens that their parents were?

Twenty-three years later Ehrlich, Holm, and Parnell (1974) could only write that

many butterflies have spring generations that are smaller and darker than their summer generations, the difference presumably being due to the seasonal variation in the environment. However, in some northern parts of their range, [they] have only a single summer generation, which is small and dark and resembles the spring generation of southern localities. In the northern populations, the individuals are presumed to have genotypes that produce the dwarfing and darkening. *Although the critical transfer experiments have not been done*, the greater constancy of the northern forms in the face of environmental changes supports these presumptions [emphasis added].

The same problem was recognized in

plants as far back as the 1920s, in the classic work of Turesson (1922, 1925, 1929) later brilliantly expanded by Clausen, Keck, and Hiesey (1940, 1947, 1948, and other papers). This work firmly established the concept of the ecotype in plant ecology and genetics, a concept more or less readily generalizable to animals in cases like those discussed by Klots and Ehrlich et al. Turesson and Clausen et al. were able, by transplant experiments, to separate phenotypic variation produced directly by the physical environment from that produced indirectly through the selection of climatically adapted genotypes. This paper is the second of a series reporting on analogous studies of North American Lepidoptera.

THE SUBSPECIES OF *POLITES SABULETI*

Situations of the sort described above are not limited to populations separated by latitude; many Lepidopterans—like the plants studied by Clausen and his colleagues—have *altitudinal* variants, often described taxonomically at the subspecies level, and these are especially interesting because of the short ground distances between the high- and low-elevation populations and the possibility of investigating the nature of their contacts, if any.

Polites sabuleti Boisduval is a small, largely tawny skipper (Hesperiiidae), widely distributed in western North America. Three named subspecies occur in California: *P. s. sabuleti*, *P. s. tecumseh* Grinnell, and *P. s. chusca* Edwards. The last is a very pale desert population and has not been examined in this study. *P. s. sabuleti* and *P. s. tecumseh* are parapatric in northern and central California, occurring at low and high elevations respectively.

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P. s. sabuleti is very widespread on sandy soils, in saline and alkaline marshlands, and in urban vacant lots. It is usually closely associated with its normal larval host, alkali grass (*Distichlis spicata* [L.] Greene, Gramineae), but may breed occasionally on Bermuda grass (*Cynodon dactylon* [L.] Pers.), an introduced lawn grass and weed. *P. s. sabuleti* is strongly multivoltine at sea level, with possibly as many as five generations a year. The flight season is very long, ranging from late March-April to mid- or late November at Sacramento and Suisun City, California.

P. s. tecumseh occurs in subalpine meadows that become dry in summer, and in alpine fell-fields in the high Sierra Nevada. Its host plant is not known, but many collectors have noted an association with species of sedges (*Carex*, Cyperaceae). Tilden (1959) reports *P. s. tecumseh* flying from July to September in Yosemite National Park, which he interprets as indicating two broods. The more complete data given in Garth and Tilden (1963) do not support this interpretation, as there is no wide spread of dates within a given year at a single locality. Farther north, at Donner Pass (7,000 feet) there is no indication of more than one brood. Emmel and Emmel (1962) found it there from 19 June to 19 August, 1960; Shapiro found it at the same localities from 11 July to 24 August, 1973, and 18 July to 12 August, 1974. The condition of Donner Pass specimens does not suggest even a partial second brood. Like many montane butterflies, *P. s. tecumseh* emerges later at higher elevations; thus, at 10,000 feet it flies mainly in August and into very early September.

As with the altitudinal subspecies of *Phyciodes campestris* Behr (Nymphalidae) previously studied (Shapiro, 1975a), those of *Polites sabuleti* are separated by a zone in which neither seems to occur. At the latitude of Sacramento, *P. s. sabuleti* is unknown as a breeding resident above 1,500 feet and *P. s. tecumseh* is unrecorded below 5,000 feet. Tilden (1959) confuses the matter by indicating that *P. s. sabuleti* extends much higher at Yosemite, but his data (given in Garth and Tilden, 1963) make it plain that this refers to the arid east slope only: the records are from Bridgeport (6,743 ft.) and Mono Lake (6,419 ft.). On the west slope

the lowest Yosemite-area record of *P. s. tecumseh* is Gin Flat (7,036 ft.) and there are no records of *P. s. sabuleti* at all (although it is abundant on the floor of the San Joaquin Valley). The nature of the east-slope contacts has not been studied, but few habitats suitable for either subspecies occur on the abrupt Sierran escarpment.

Polites sabuleti tecumseh differs from summer *P. s. sabuleti* in being smaller, hairier, and more heavily marked, especially on the hindwing ventrally. The dark markings on this wing are often described as being a "colder," grayer color than in *P. s. sabuleti*. Although a series of 23 *tecumseh* from the vicinity of Donner Pass shows considerable variation, no seasonal pattern is apparent. In *P. s. sabuleti* from Sacramento and Suisun City (over 450 specimens examined) there is marked seasonal variation: March-May and September-November specimens are, on the average, smaller, darker, and hairier than summer ones, and some are superficially exceedingly similar to *P. s. tecumseh*, although there are minor (but fairly consistent) differences in certain details of the pattern. The phenotypes of wild specimens of both taxa are illustrated in Figures 1-3.

EXPERIMENTAL METHODS AND RESULTS

Would stock of *P. s. tecumseh* reared under conditions that produce summer phenotypes of *P. s. sabuleti* produce the normal *tecumseh* phenotype, or would it be modified in the direction of the lowland, summer one? Ova were obtained from a female *tecumseh* collected at Donner Pass (7,000 ft.), 24 July 1974, and from two female *sabuleti* collected in a salt marsh at Suisun City, Solano County (10 ft.), 6 August 1974. The resulting progeny were reared side by side in plastic Petri dishes (5½" diameter X ¾") at comparable densities (5-8 larvae/dish) under continuous illumination from a 60w bulb at 25C (77F). All larvae were fed fresh cuttings of Bermuda grass (*Cynodon dactylon*), and mortality in both stocks was negligible. Thirty adult *P. s. sabuleti* (16 ♂ 14 ♀) and thirteen *P. s. tecumseh* (8 ♂ 5 ♀) were obtained. Continuous light was selected as a regime ecologically insignificant to both stocks but known



Fig. 1. Dorsal and ventral surfaces of summer specimens of *Polites sabuleti sabuleti* from the Central Valley of California.



Fig. 2. Dorsal and ventral surfaces of early spring and late fall *P. s. sabuleti* from the Central Valley, approaching the phenotype of *P. s. tecumseh*.



Fig. 3. Dorsal and ventral surfaces of *P. s. tecumseh* from Donner Pass, California (7,000 feet), July-August.

to inhibit diapause in *P. sabuleti* and related species.

The two stocks differed in several respects in the laboratory. First- and second-instar larvae of *P. s. sabuleti* were yellowish green; in the third instar they turned purplish brown; and thereafter they remained that color. *P. s. tecumseh* larvae were purplish brown throughout their development. At corresponding points in the life cycle the early stages of *P. s. sabuleti* were always larger than their high-altitude counterparts. The developmental rates of the two stocks differed very significantly, with little overlap: from egg to adult *P. s. sabuleti* took 39-76 days (weighted mean, 58.5 days) and *P. s. tecumseh*, 70-111 days (weighted mean, 86.0). No diapause was observed in either

stock. Normally, high-elevation or -latitude stocks of Lepidoptera develop more rapidly than conspecific ones from more temperate climates when reared under uniform laboratory conditions; the reversal of this situation in *Polites sabuleti* is to my knowledge unique in Lepidopteran stocks in which diapause is not manifested in culture.

The developmental differences noted above were not mirrored in larval behavior or morphology. The adults, however, were obviously different and "true" to their normal phenotypes (Figs. 4, 5): nondiapaused *tecumseh* reared at high temperatures retained all of their distinguishing characters, including size. It thus appears that the complex of characters present as a developmental option in low-



Fig. 4. Dorsal and ventral surfaces of representative bred *Polites sabuleti sabuleti*; continuous light, 25C.

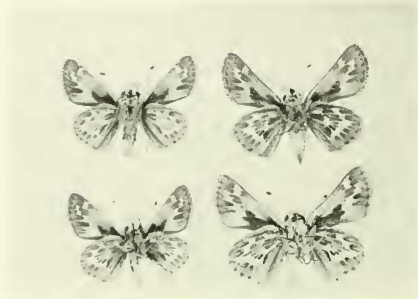
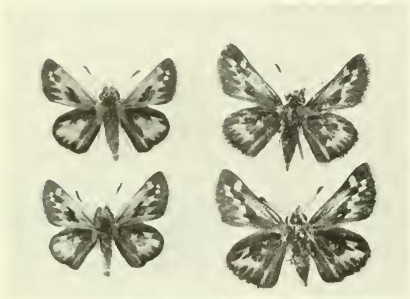


Fig. 5. Dorsal and ventral surfaces of representative bred *P. s. tecumseh*; same conditions as in Figure 4.

land, multivoltine populations is genetically fixed in *P. s. tecumseh*, confirming Ehrlich et al.'s prediction.

DISCUSSION

Three sets of "altitudinal subspecies" have now been investigated in butterflies, representing three different and quite unrelated families. They are *Pieris occidentalis* Reakirt and its alpine representative, familiarly (but incorrectly) known as *P. o. "calyce"* Edwards (Pieridae); *Phyciodes campestris* Behr and its montane subspecies *montana* Behr (Nymphalidae); and *Polites sabuleti*. The first two are discussed at length in Shapiro (1975a). Each species presents a picture different from the others.

Pieris occidentalis shows very little, if any, genetic differentiation of the univoltine, monophenic and bivoltine, diphenic populations of high and moderate elevations, respectively. The alpine stock retains the ability to produce an estival phenotype and to develop without diapause, and its own phenotype is indistinguishable from the vernal one produced downslope. Their mating behavior involves male aggregations on mountain-tops, a behavior pattern conducive to gene flow, and in laboratory experiments no reproductive barriers have been found between uni- and bivoltine populations.

Phyciodes campestris shows a superficially similar picture, in that the high altitude subspecies *montana*, when reared under outdoor conditions at sea level, produces the foothill phenotype rather than its own. However, the cold-season phenotype of lowland populations is quite different from *montana*, and the ability to produce the *montana* phenotype appears to be restricted to high-elevation populations. In this case, then, phenotypic plasticity is not reciprocal, and the high-elevation population is genetically differentiated.

Polites sabuleti shows the highest degree of differentiation yet encountered. The possibility that it has achieved reproductive isolation (i.e., speciation) between high- and low-elevation populations cannot be discounted. Because of the difference in developmental time, no crosses between the stocks could be made. They are not known to intergrade anywhere, since the altitudinal discontinuity between

them seems to run the length of the Sierra Nevada.

These experiments have been duplicated with a latitudinal subspecies pair—California *Pieris occidentalis* and its subspecies *P. o. nelsoni* Edwards from Fairbanks, Alaska (Shapiro, 1975b). Their phenotypic differences are clearly heritable, and the response of both phenotype and diapause to photoperiod has been observed in F_1 and F_2 hybrids.

Studies of the *Pieris occidentalis* and *P. napi* L. species complexes (Shapiro, 1975c) strongly imply that univoltinism is evolutionarily derivative from multivoltinism, accompanying the successful invasion of increasingly rigorous climates. High-altitude and -latitude populations of widespread species are probably derived from lowland sources, as has been well documented for the Sierran alpine flora (Chabot and Billings, 1972). The overall picture emerging from these studies supports the suggestion that seasonal phenotypes of multivoltine populations may become fixed through selection of modifiers influencing thresholds of developmental expression ("genetic assimilation," Waddington, 1953). The somewhat unusual circumstances in *Phyciodes campestris montana* will be explored in another paper.

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